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Effect of Non-Genetic Factors on Semen Quality of Cocks and Repeatability of Estimates

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ABSTRACT

Semen quality determines the fertilizing ability of cocks and other male animal's semen. The goal of this study was to ascertain how ecotype, collection week, cock, and their interactions affected semen quality. The study also estimated the repeatability of some semen quality traits. A total of forty (40) cocks made up of 20 heavy ecotypes and 20 light ecotypes were used for the study. The cocks were individually identified with wing tags. Semen was collected from each cock from 7 am to 10 am every Saturday consistently for six weeks using the abdominal massage technique (AMT). Collected semen was evaluated in terms of volume, concentration, mass density, live sperm, and pH. The results indicated that the week of semen collection significantly affected sperm motility, percent live sperm, and semen pH, while ecotype significantly affected live sperm only. Ecotype x week x cock, interactions significantly affected sperm concentration only. The repeatability of semen quality traits was generally low and ranged from 0.01 ± 0.04 to 0.12 ± 0.04, thus depicting the massive influence of environmental factors on semen quality traits. It is concluded that more than six records are required to characterize the semen-producing ability of the Nigerian indigenous cocks.

Keywords: Cock, Ecotype, Producing ability, Repeatability, Semen quality

INTRODUCTION

Semen quality determines the fertility of cocks when their semen is inseminated in hens or through natural mating. Therefore, semen quality plays a crucial part in the economy of poultry production. Both genetic and non-genetic factors have been noted to affect semen quality. Ayeneshet et al. (2024) reported that ejaculate volume, semen pH, and sperm concentration were affected by breed, age, individual cocks, season, collection frequency, light, and other environmental factors. According to Ordaz-Contreras et al. (2023), ejaculate volume, Progressive motility, and sperm concentration decreased significantly due to age. Still, semen concentration did not differ with seasons and collection time. According to Mavi et al. (2019) and Ayeneshet et al. (2024), there are discrepancies between the semen quality of domestic and exotic cocks. Sitanggang et al. (2020) observed that the age of bulls, the collection season, the frequency of ejaculation, and the collection interval affect semen quality. The fertilizing ability of semen can be accessed by measuring motility, live/dead sperm, and morphological abnormalities (Okoro et al. 2016). The coefficient of repeatability describes the degree of similarity between consecutive measurements affected by genetic and random environmental factors (Pelayo et al., 2019). Sanda et al. (2022) state that repetition is crucial to the poultry industry's profitability and that the magnitude of repeatability estimates indicates how much selection at any stage would impact flock performance later on. Nonetheless, this will assist breeders in arranging the best possible combination for the highest possible financial gain. Repeatability is also crucial when measuring an animal's capacity to produce offspring and determining how selection has affected a population's performance (Akporhuarho and Obodoagwu, 2020). It is employed to determine a stock's transferability, reliability of performance or behavior across time, and ability to maintain performance and position within a trial group on subsequent generations. To establish repeatable estimates of semen volume, sperm motility, sperm concentration, percent live sperm, and semen pH, this study set out to ascertain the effects of ecotype, week of collection, cock, and their interaction on semen quality.

MATERIALS AND METHODS

Ethical Approval

The Department of Animal Science, Delta State University Animal Care and Use Committee approved the protocol for the conduct of the experiment (Approval NO 2021-18).

Study Location

The poultry unit of the teaching and research farm, Department of Animal Science, Delta State University, Asaba Campus, Asaba, Nigeria, is where the study was carried out.

The Experimental Cocks

In total, 40 indigenous cocks comprising 20 heavy and 20 light ecotype types aged 26 to 28 weeks were used. The average weight of the heavy and light ecotype cocks were 1.60kg and 0.96kg, respectively. The heavy and light ecotype cocks were raised separately in deep litter pens from day-old to sexual maturity. The researchers prepared the feed and water *ad libitum* throughout the study. The broiler finisher ratio was used because of its high crude protein of 20% and high metabolizable energy value of 3.3 ME/kg. Throughout the research, there was no fatality.

Semen Collection

Each cock was wing tagged with a number and trained for artificial ejaculation through the abdominal massage technique (AMT) for 14 days before the commencement of the trials. The training was necessary because the cocks were not used to massage and artificial ejaculation before. Semen was collected from 7am to 10 am on Saturdays for 6 weeks using the AMT described by Burrows and Quinn (1937). Semen was collected into sterile test tubes. The average daily semen volume was 0.12 ± 0.02 (heavy ecotype) and 0.14 ± 0.04 (light ecotype). The semen concentration per ml was 182.50 x 106 ± 13.49 and 171.43 x10^6 ± 22.76 for heavy and light ecotypes, respectively.

Semen Analysis

The semen was evaluated immediately after collection for sperm concentration and motility. Other characteristics, such as volume, pH, color, mass activity, live-dead ratio, and morphological abnormalities, were also examined. The volume of freshly ejaculated semen was determined using a syringe of 1 ml. Semen color was evaluated by sight to be milky, creamy, or transparent. Semen pH was determined using a pH indicator paper. The motility of the semen was determined immediately. On a glass slide, a drop of semen was deposited, covered with a cover slip, and examined under an Olympus microscope (Japan) at x 40 magnification. The percentage motility was scored visually. Semen concentration was determined using a Neubauer chamber haemocytometer with a coverslip. The ratio of normal saline to the semen was 1:400. The haemocytometer was filled with the diluted semen using the red pipette and counted under the microscope. The mass activity was determined immediately after semen collection as the speed at which the sperm cells traveled across the microscopic field. According to Akintunde et al. (2020), the mass activities were graded as 0 = no mass activity, + = slow wave motion, ++ = rapid wave motion, and +++ = very rapid wave motion. The Live and dead ratio was determined using the nigrosin/eosin staining technique adopted by (Adejoh-Ubani et al., 2022). When viewed under a microscope, the sperm cells that did not absorb the stain were recorded as live, while those that absorbed the stain were counted as dead.

Statistical Analysis

The following is a description of the statistical model that was used to analyze the data: $Y_{ijkl} = Ecotype_i + Week_j + Cock_k + Ecotype_i \times Week_j + Ecotype_i \times Cock_k + Week_j \times Cock_k + Ecotype_i \times Week_j \times Cock_k + E_{ijkl.}$ Where Y_{ijkl} is the observation of semen trait, e_{ijkl} is the residual. The restricted maximum likelihood (REML) method of the lme4 R package (R version 64. 3: 1: 3) was used to obtain the variance components necessary for the repeatability estimation. The fixed effect of ecotype, week, cock, and their interactions on the semen traits was tested using the function of repeatability (R).

The standard error of repeatability was calculated using the standard expression given by Becker (1984).

RESULTS AND DISCUSSION

The effects of ecotype, week, cock, and their interactions on the quality of the semen are shown by the mean square values in Table 1. Live sperm (LS) and sperm motility (SM) were both significantly affected by semen collection week ($p < 0.01$). The effect of ecotype was significant ($p<0.05$) on live sperm, while ecotype x week x cock interaction was significant ($p<0.01$) on sperm concentration.

Table 1. Mean square values for the effect of ecotype, week, cock, and their interactions on the semen quality of Nigerian Indigenous cocks

Traits	E (1)	W (1)	C (1)	E x W (1)	Ex C (1)	W x C (1)	ExWxC (1)	Residual (37)
Vol.	0.038	0.075^{ns}	0.049	0.007	0.005^{ns}	0.008	0.000	0.024
Con.	3664	37618^{ns}	18471^{ns}	11532^{ns}	1115	1009	125379^{**}	11043
Mot.	1017.6^{ns}	7200^{**}	1269	33.3	248.3	207.7^{ns}	1976.3^{ns}	677.0
LSp	2631.7^{*}	8526^{**}	298	0.00^{ns}	316.8	313.9^{ns}	1269^{ns}	472.0
pH	0.0023^{ns}	29.47^{**}	0.02^{ns}	0.867^{ns}	3.682^{ns}	4.770^{ns}	5.741^{ns}	3.493

Note: * p<0.05,** p<0.01, ns: not significant (p>0.05). Degrees of freedom for sources of variations are in parentheses. Vol = Volume, Con = Concentration, Mot = Motility, LSp = Livesperm, pH = semen pH. E = Ecotype, W = Week, C = Cock.

Table 2 shows the impact of semen collection week on the quality of cocks' semen. With a rise in weeks, the percentage of living sperm increases dramatically (p<0.01). Percent live sperm increases significantly (p<0.01) with an increase in weeks. This implies that the higher the week of semen collection, the higher the percentage of live sperm.

Motility followed a similar trend with live sperm as the percentage motility of sperm cells increased significantly (p<0.01) within the week of collection. This implies that those factors increase semen viability within the week of collection. To boost fertility, cocks used for artificial insemination should be trained for artificial ejaculation for a more extended period before using their semen, according to the results of the influence of weeks on live sperm and motility. The mean pH of semen collected in week 1 was significantly (p<0.01) lower than the pH value of semen, which ranged from 7.92-7.96, as reported by Emeka et al. (2022). However, the mean pH values of semen collected from weeks 2 to 5 did not vary significantly (p>0.01) from each other and were within the optimum pH value of cock semen. The mass activity was +++ on average, implying very rapid wave motion.

Table 2. Effect of week of collection on the semen quality of Nigerian ecotype cocks

Traits	Week 1	Week 2	Week 3	Week 4	Week 5
Live sperm	35.78 ± 4.13^{b}	51.11 ± 3.43^{ab}	59.56 ± 3.02^{ab}	65.11 ± 3.66^{ab}	77.44 ± 3.16^{ab}
Motility	43.33 ± 5.32^{b}	56.11 ± 4.10^{ab}	62.00 ± 3.16^{ab}	65.56 ± 3.84^{ab}	83.33 ± 3.32^{b}
pH	5.00 ± 0.56^{b}	7.39 ± 0.10^{a}	7.67 ± 0.07^{a}	7.78 ± 0.07^{a}	7.68 ± 0.07^{a}

Mean values along the same row with different superscript letters are significantly (p<0.01) different.

Table 3 shows that there is a significant (p<0.05) difference in the effect of ecotype on the live sperm of cocks. In contrast, the motility between heavy and light ecotype cocks was not significantly different (P>0.05).

Table 3. Effect of ecotype on the live sperm of Nigerian Indigenous cock semen

Ecotype	Live sperm (%)	Motility (%)
1	64.64^{a}	71.21
2	49.25^{b}	69.50

a>b (p<0.05)

The heavy ecotype (ecotype 1) produced sperm cells that have the highest percentage of live sperm (64.64%) than the light ecotype (ecotype 2) with 49.25%, as shown in Table 3. This implies that the heavy ecotype cocks can sire more progenies than the light ecotype. The values for live sperm and motility are inconsistent with previous reports (Mavi et al., 2018; Bhatti et al., 2023; Ayeneshet et al., 2024). Table 4 shows the semen quality of cocks' correlation coefficients between cocks, within cocks (residual), and class.

Table 4. The between cocks, within cocks, and intraclass correlation coefficients of semen traits of Nigerian ecotype cocks

Traits	Between cocks	Within cocks	Intraclass correlation
Volume	0.002	0.02	0.12±0.05
Concentration	175.80	14419.90	0.01±0.04
Motility	30.95	656.12	0.05±0.06
Live sperm	43.36	1286.58	0.03±0.05
pH	0.16	3.39	0.05±0.06

The repeatability estimates of semen volume (SV), sperm concentration (SC), sperm motility (SM), live sperm (LS), and semen Ph (SPH) were very low, ranging from 0.01 ± 0.04 to 0.12 ± 0.05. This implies that other nongenetic factors, such as environmental temperature, may have interfered on different occasions during semen collection and analysis. Udeh (2010) reported low to moderate repeatability estimates of 0.43, 0.31, 0.03, and 0.02 for semen volume, sperm concentration, progressive motility, and percent live sperm, respectively, of Nigerian Indigenous cock's semen using 8 records. It is also in line with the report of Kassa (2018) that environmental influences significantly influence reproductive qualities in hens and that these traits have poor heritability estimates. In contrast, Emeka et al. (2022) reported repeatability estimates of 0.50, 0.56, 0.50, 0.55, 0.39, 0.38, and 0.42 for semen volume, semen Ph, sperm Concentration, normal morphology, active motility, live sperm, and viability respectively, which are higher than the estimates obtained in this study.

CONCLUSION

It was concluded that the week of semen collection, ecotype, cock, and their interactions affected some semen traits. The low repeatability obtained implies that more than six records are required to characterize the semen-producing ability of Nigerian Indigenous cock.

Conflicts interests

There was no conflicting interest between the authors or any other person.

Ethical Consideration

The authors checked ethical issues relating to plagiarism, data fabrication and falsification, double publication, and/or submission.

Author contributions

Ifeanyichukwu Udeh designed and supervised the research project. Ifeanyichukwu Udeh, Sorhue Godstime Ufuoma, Joy Afokoghene Edegbo, and Moemeka Mike Adimabua did semen collection and evaluation. Ifeanyichukwu Udeh did data analysis. Joy Afokoghene Edegbo did the write-up, Ifeanyichukwu Udeh revised it, and Sorhue Ufuoma Godstime prepared the manuscript for publication.

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